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# MICROBIAL PROFILE OF LAMINAR FLOW CLEAN ROOMS

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LAMINAR FLOW CLEAN ROOMS

by  
Edmund M. Powers, M. S.  
Space Biology Branch  
Laboratory for Atmospheric  
and Biological Sciences

September 1965

Goddard Space Flight Center  
Greenbelt, Md.

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### SUMMARY

The microbial contamination in the air and on surfaces of spacecraft and table tops during exposure in laminar downflow and crossflow rooms was evaluated. The concentration of viable particles was extremely low and well within the requirements specified by NASA.

The concentration of viable particles in the air of the two rooms over a 4-hour period was a maximum of 0.04 viable particles/ft<sup>3</sup> of air in the occupied downflow room and 0.5 viable particles/ft<sup>3</sup> of air in the occupied crossflow room. The NASA standard for air of bioclean rooms is 2.0 viable particles/ft<sup>3</sup>. The counts were approximately the same in the air of both clean rooms when the rooms were unoccupied and the airflow was on. Personnel did not appear to contribute significantly to the microbial contamination in the air of the downflow room, but counts were increased significantly by personnel in the crossflow room both upstream and downstream of the sampling devices. The level of contamination in the air downstream of personnel was not significantly higher than the air upstream of personnel in the crossflow room.

The viable contamination on the surface of the nosecone, landing capsule, and table top was measured by the Rodac impression technique. Counts on these surfaces ranged from 0 to 23 viable particles/ft<sup>2</sup> in the downflow room and from 0 to 58 viable particles/ft<sup>2</sup> in the crossflow room. The viable contamination on all three surfaces over a 4-hour period did not appear to be significantly increased when personnel were introduced into the downflow room, but in the crossflow room, counts on these same surfaces were increased by the presence of personnel, particularly when personnel worked upstream of them.

Handling of a landing capsule with gloved hands did not significantly increase the microbial contamination on its surface in either clean room. This result indicates that the use of sterile clothing, including gloves, and proper handling procedures together with a laminar airflow could reduce microbial contamination considerably.

The efficiency of the laminar flow room in providing clean air and the extremely low counts obtained, particularly in the downflow room, have considerable

significance for spacecraft assembly and sterilization. By reducing contamination to a minimum, sterilization times might be reduced and sterilization procedures made less harsh. The small degree of contamination obtained in laminar flow clean rooms might also insure that our sterilization procedures are effective.

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## MICROBIAL PROFILE OF LAMINAR FLOW CLEAN ROOMS

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### INTRODUCTION

The microbial profile of two types of laminar flow clean rooms was evaluated by Goddard Space Flight Center in cooperation with the Sandia Corporation, contractor for the U.S. Atomic Energy Commission, Albuquerque, New Mexico.

A search of the literature reveals that very little is known about the viable microbial contamination in clean rooms, particularly that in laminar flow clean rooms. The use of clean rooms for spacecraft assembly makes it essential to know what levels of microbial contamination can be expected under varying working conditions and to set limits for the viable contamination of air and surfaces in clean rooms.

Hobby estimated (ref. 1) that the microbial population of the surface of a spacecraft is in the order of  $10^9$  microorganisms per 100 square feet. Calculations showed that 22.1 hours at  $135^\circ\text{C}$  would be required to reduce the population so that there would be 1 chance in 10,000 of a single organism surviving. Therefore, to achieve a probability of not more than 1 chance in 10,000 of landing a viable particle on a planet, microbial contamination on the spacecraft must be held at the lowest possible level so that an acceptable degree of confidence in the effectiveness of the terminal heat sterilization cycle is obtained (ref. 1). Contamination control procedures must be established, improved upon, and closely monitored to verify their adequacy. Assembly operations during assembly and test of actual flight hardware must also be extensively monitored.

Because these monitoring procedures vary considerably among investigators, a great need exists to standardize them so that results of clean rooms may be compared and so that some measure of confidence in clean rooms in various parts of the country may be obtained.

The methods commonly used for sampling air include impaction on solid media (settling plates and strips) and impingement in liquid media. Surfaces are usually sampled with swabs or contact (Rodac) plates. Goddard (ref. 2) listed the advantages and disadvantages of several methods used for sampling air and surfaces and proposed an excellent method for sampling the microbial contamination of surfaces using Rodac plates.



Danielet al. (ref. 1), who developed criteria for an assembly, test, and sterilization facility for Jet Propulsion Laboratory (JPL), compared air sterilization equipment and concluded that a combination of air filtration and air washing offered the most economical and effective arrangement to meet basic air cleanliness requirements. In this connection, the Whitfield (refs. 3 and 4) laminar air flow system offered excellent possibilities for the assembly of spacecraft and for a clean room and sterilization facility.

The laminar air flow system when in combination with adequate air filtering systems has great potential for increasing clean-room standards of cleanliness by several orders of magnitude. The laminar concept developed by Sandia Corporation (ref. 3) uses a single pass technique in which one wall or ceiling consists of a complete bank of high-efficiency particulate air (HEPA) filters. These filters collect 99.97 percent of all contamination 0.3 micron in size and larger before it enters the clean room. Because of their resistance these filters tend to create a uniform air flow throughout the room. The opposite wall or grated floor is designed as a complete return air plenum; the air makes a single pass across the room either in a horizontal (laminar crossflow room) or downward (laminar downflow room) flow pattern. The laminar flow technique is designed for as high as 800 air changes per hour while the conventional type clean room is normally designed for only 20 air changes per hour (ref. 5).

Daniel et al. (ref. 1) evaluated both laminar downflow and laminar crossflow systems and concluded that the laminar downflow system offered the best solution for the design of an ultraclean facility. Economically, the per unit cost of a laminar crossflow room may be less than the cost of a laminar downflow room, all factors being equal; however, by decreasing the air velocity to a permissible level and increasing the size of the room, the per unit cost of the laminar downflow concept should approach the cost of a comparable laminar crossflow installation since the amount of equipment and number of personnel can be increased, reducing the unit cost per square foot of working space.

Portner (ref. 6) studied the level of microbial contamination in a Martin Company clean room (conventional type) over a 52-week period. She found that the number of microorganisms per cubic foot of air was about 10 times greater in the factory than in the clean room and that aerobes appeared to be more prominent than anaerobes. She also reported that the microbial contamination on a stainless steel surface due to aerial fallout rapidly reached a maximum level and remained more or less constant throughout the year. This study also points out the need for clean room facilities.

The present study was undertaken to determine the microbial contamination in the air and on surfaces in laminar downflow and laminar crossflow rooms.

Because of its configuration, a full scale conceptual model of a planetary lander vehicle and its protective nosecone were also monitored to determine the microbial contamination on its surface during exposure in these laminar flow clean rooms. The results, herein reported, indicated that the laminar flow clean room is extremely effective in controlling microbiological contamination and that this system far surpasses existing requirements for bioclean room facilities. The use of the laminar downflow room for spacecraft assembly has great potential and should be seriously considered.

## MATERIALS AND METHODS

### Laminar Downflow Clean Room

The clean room in which these tests were performed is located at Sandia Corporation in Albuquerque, New Mexico, and is composed of three separate rooms connected to form a single unit. Each end room is approximately 8 ft by 10 ft. The center section, in which all experiments were performed, is approximately 10 ft by 20 ft. Each of the three sections has its own air-handling/air-conditioning unit; 4-foot sliding doors at the center of each end of each unit provide free access through the entire test area. The ceiling of each room consists of fiberglass high-efficiency particulate air (HEPA) filters through which air enters. Fluorescent lights having little air resistance run the length of the room. The floor is a standard, heavy-duty bar-grating through which the air exits from the room. All sections are constructed of stainless steel to facilitate cleaning (ref. 7).

The air flows vertically downward through the entire ceiling at 100 linear ft per minute in the main room (approximately 800 air changes per hour). Temperature was controlled at  $70^{\circ}\text{C} \pm 2^{\circ}$  and humidity to  $30 \pm 10$  percent. All filters and filter mountings were checked thoroughly for penetration using smoke, and all leaks were sealed. Subsequent tests for airborne particulate contamination with a Royco particle counter (PC-200-A) confirmed that there was zero penetration of the filters by smoke particles (size range averaging about 0.25 micron). Airborne particle count (0.3 micron and larger size) was zero throughout the room, providing there was no particle generation between the filter and the counter (ref. 7).

Before this study began the room was vacuumed and all surfaces were washed with a mild detergent.

### Laminar Crossflow Clean Room

The clean room in which this study was performed is located at the Gulton Industries, Albuquerque, New Mexico. The air flow of the crossflow room differs

somewhat from the air flow of the downflow room in that it is directed horizontally across the room from one wall to the opposite wall. The two walls consist of filter banks, with rough prefilters in the wall at the air-exhaust end of the room, and final absolute filters in the opposite wall at the air-input end of the room. The absolute filters are fiberglass high-efficiency particulate air (HEPA) filters, which filtered out all particles 0.3 micron and larger from air entering the room. The remaining two walls and ceiling are of dry wall construction and painted with vinyl paint. The floor is solid and covered with vinyl tile.

This room meets the specifications of Federal Standard 209 (ref. 8) for particulate matter and is rated as class 100 at the air-input end and class 10,000 or better at the air-exhaust end of the room. The dimensions of the room are 33 ft wide, 96 ft long, and 9 ft high. Because of the length of the crossflow room, the rate of airflow was 130 ft per minute. Temperature was controlled at  $22^{\circ}\text{C} \pm 2^{\circ}$  and humidity to 35 percent.

Tests for leaks in the filters and filter mountings and for airborne particulate contamination were conducted about 6 months before this study was initiated. These tests were performed by the same methods used in the downflow room.

This crossflow room is used by the Gulton Industries for the assembly of electronic circuits and is routinely occupied by 20 women and 3 to 5 men. The entrance and exit to the room is located at the exhaust end.

#### Mars Spacecraft

Figures 1 and 2 show the general configuration of the Mars landing capsule and the protective nose cone respectively. Figure 3 is a photograph of the landing capsule and nose cone as they appeared in the clean room during the sampling procedure. The capsule is 23 inches long by 15 inches high; the nosecone has a diameter of 16-1/2 inches and is 27 inches long. A technical description of the spacecraft can be found in the Planetary Project Document (ref. 9).

The nosecone and capsule were placed side by side on a grated table 12 inches high to permit easy access to the interior of the cone (Figure 3). The nosecone was divided into four parts: the exterior aluminum surface into two equal halves and the interior fiberglass surface into two equal halves. Each of the four parts was gridded to form 25 equal squares. The grid intersections were numbered 1 through 16 in a serpentine fashion; at each sampling sequence, 32 Rodac impression samples of the cone were taken by sampling the 8 odd (red) intersections of each of the four parts at one sampling sequence and the even (black) intersections at the next sampling sequence.

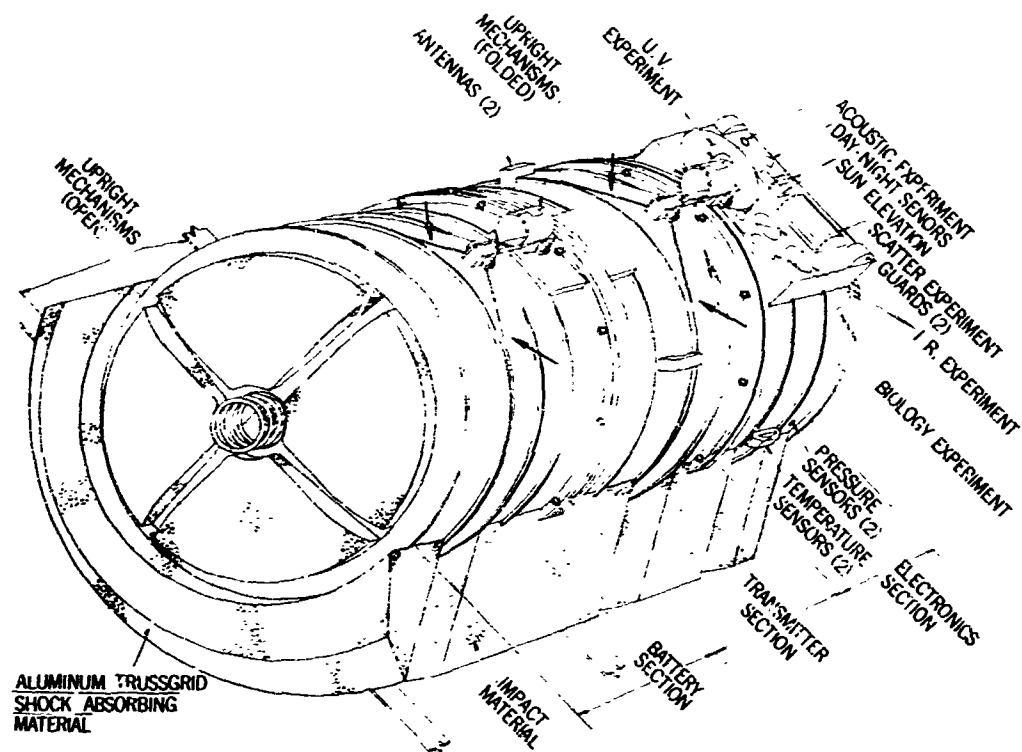


Figure 1. Mars Landing Capsule Configuration

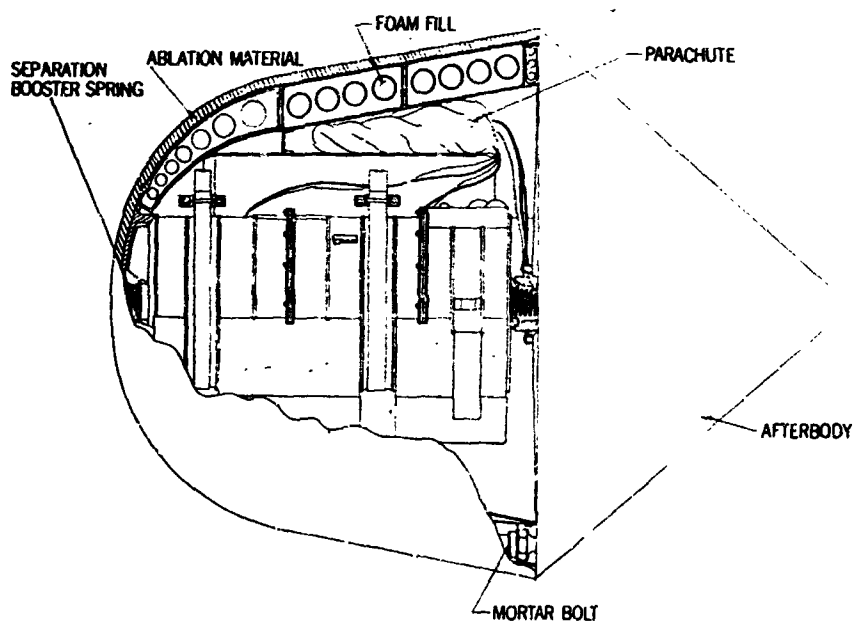
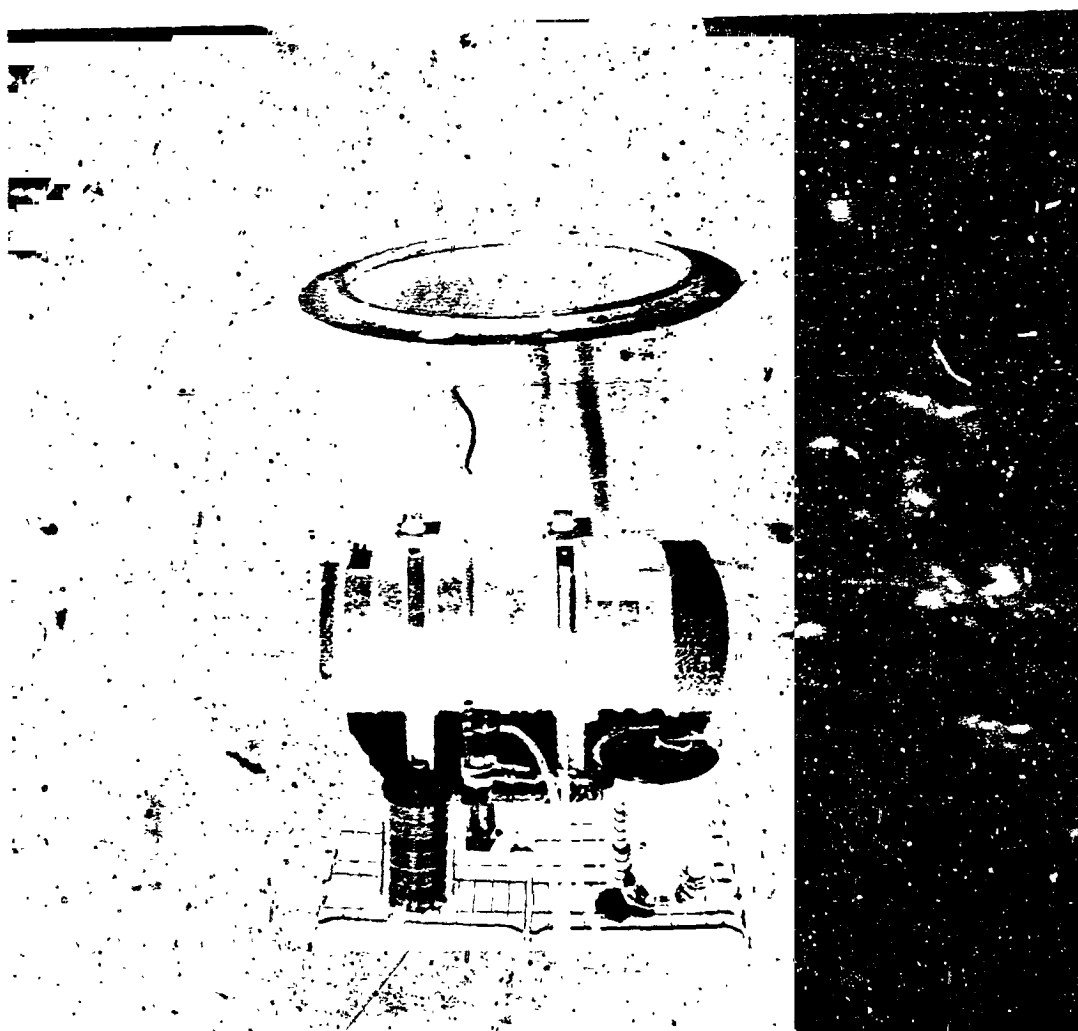


Figure 2. Protective Nosecone Configuration



**Figure 3. Photograph of Mars Landing Capsule and Protective Nosecone**

The landing capsule, which was not gridded, was placed on the table with the metal surface up. The bottom of the capsule was covered by a trussgrid shock-absorbing material, which was not sampled for microbial contamination. Rodac impression samples of the capsule were taken from the flat metal straps which surround the capsule and serve as upright mechanisms when

released and from the flat surface at one end and on top of the capsule (Figure 3). Ten Rodac samples were taken at each sampling sequence.

In the downflow room the spacecraft were placed in the center of the room. In the crossflow room the spacecraft were placed downstream of personnel and approximately 10 ft in front of the air outlet for one sampling sequence and upstream of personnel and approximately 6 ft in front of the air supply for the next; when the room was unoccupied, the spacecraft were placed in the center of the room.

The cone and the capsule were decontaminated with 80 percent isopropyl alcohol 10 to 15 minutes before the first surface samples were taken at zero hour.

#### Air Sampling

Andersen samplers (ref. 10), which are six-stage sieve type samplers, were used to collect viable airborne particles in the clean rooms. Each stage contains a perforated plate with 400 holes immediately below which was a plate of Trypticase Soy Agar (BBL).

Four Andersen samplers were employed simultaneously for each sampling sequence. They were positioned in the downflow room as follows: (1) in front of the entrance, 3 ft above the floor; (2) in the middle of the room, approximately 2 ft from the spacecraft and 7 ft above the floor (1 ft below the air supply); (3) on the grated table next to the landing capsule; (4) at the far end of the room, 1 ft above the floor.

In the crossflow room, three of the Andersen samplers were placed at benchtop level and one was placed on the grated table with the spacecraft. All samplers were placed so that they were downstream of all workers for one sampling sequence and upstream for the next sampling sequence. When the room was unoccupied, the samplers were positioned around the spacecraft in the center of the room.

Each sampler was sterilized by wrapping in paper and autoclaving at 121°C and 15 lbs pressure for 30 minutes. Petri plates were wiped with 80 percent isopropyl alcohol and allowed to dry before being loaded into the Andersen sampler.

Air was drawn through the samplers with 1/6-hp vacuum pumps at a rate of 1 cubic foot per min (cfm) for 15 minutes. Flowmeters were used to measure the airflow through each device before sampling. The vacuum pumps were placed on the floor so that any fumes that might have escaped were immediately carried out of the room by the airstream.

Continuous and uninterrupted sampling of the viable particles per unit volume of air was accomplished with an Andersen 0800 Monitor for airborne bacteria (ref. 11). This device consists of an agar-coated drum, 5-1/2 inches in diameter by 3-5/8 inches high, mounted on a threaded shaft in a 3-1/2 quart stainless steel beaker. The drum is rotated by a Cramer timer mounted on the cover. A jet for impacting airborne particles on the drum is mounted in the side of the beaker. A stainless steel jet calibrated to deliver 3 liters of air per minute was employed and the flow rate was adjusted with a flowmeter to the specified flow rate of the jet. The 0800 Monitor was placed on the grated table next to the nosecone. The beaker was cleaned and decontaminated with 80 percent isopropyl alcohol prior to use.

#### Surface Sampling

Rodac plates developed by Hall and Hartnett (ref. 12) were used to determine the level of microbial contamination on surfaces (spacecraft and stainless steel table) in the clean room. Each plate was filled with 15.5 to 16.0 ml of agar medium to form a meniscus above the edge of the plate which allowed impressions of the agar onto a surface. Samples were taken by impressing the agar directly on the surface to be sampled. Plates were then incubated and colonies counted directly on the agar.

#### Stainless Steel Table

A stainless steel table 6 ft long by 3-1/2 ft high by 3 ft wide was placed next to the spacecraft in both clean rooms. The table top was gridded in the same manner as the nosecone, i.e. into 25 equal squares. The intersections were numbered 1 through 16 in a serpentine fashion. The odd and even intersections (8 each) were sampled on alternate sampling periods by the Rodac impression method.

#### Microbial Fallout

Microbial fallout was determined by exposing commercially purchased blood agar plates (BBL) in three different areas of the room at bench-top level. Five plates were exposed in each area for 2- and 4-hr periods during each sampling sequence.

#### Media

Trypticase Soy Agar (BBL) was used for all sampling procedures. Media was dispensed with an automatic pipetting machine into glass petri dishes (for the Andersen samplers) in 27-ml volumes and into Rodac plates in 15.5-ml volumes. The agar drums in the 0800 Monitor received approximately 200 ml of

agar medium. All plates and agar drums were incubated at 35°C for 48 hrs prior to use to determine their sterility and to dry them sufficiently for sampling.

#### Clothing

The investigator who collected the samples wore sterile clothing consisting of cotton cap, face mask, surgical gown, and sterile rubber gloves. The clothing was donned just before entering the room. Personnel who occupied the downflow room for 2- and 4-hr periods wore only a surgical gown and personnel who occupied the crossflow room wore only a smock and booties.

#### Sampling Procedure

The downflow room and the nosecone, capsule, and stainless steel table in the room were monitored under the three following conditions:

- Airflow turned off and room unoccupied
- Airflow turned on and room unoccupied
- Airflow turned on and room occupied by two workers

When the room was unoccupied, the sampling sequence was at 0, 2, 4, 6, and 23 hr. Because the room was occupied for a maximum of 4 hr, the sampling sequence during occupancy was at 0, 2, and 4 hr.

Prior to each sampling sequence, the nosecone, capsule, and stainless steel table were decontaminated with 80 percent isopropyl alcohol. The first samples were collected approximately 15 minutes after decontamination (zero hr). No further decontamination was made during the sequence. Sterile clothing was donned and all materials necessary for sampling were prepared before the room was entered. Surface samples were taken by two workers immediately upon entering the room and sampling was accomplished in less than 5 minutes. The Andersen samplers were then placed into position and turned on for 15 minutes. The two workers left the room while the Andersen samplers were operating, except of course when the room was sampled under occupied conditions.

Sampling conditions differed slightly in the crossflow room because the air supply could not be turned off. Air and surface samples were taken in the crossflow room under the following conditions:

- Room occupied and all samples taken upstream of working personnel
- Room occupied and all samples taken downstream of working personnel
- Room unoccupied and samples collected midway between the air inlet and the air outlet

During occupancy 20 to 25 people (20 women and 3 to 5 men) were in the room at all times. Samples were collected at 0, 2, 4, and 6 hr while the room was occupied and at 0, 2, 4, 6, and 23 hr while the room was unoccupied. The 20 to 25



people who worked in the room assembling electronic circuits wore only lint-free frocks and boots.

During occupancy of both clean rooms, the capsule was sampled by the Rodac impression method before and after handling with gloved hands. Sampling was accomplished by taking ten Rodac samples of the capsule after decontamination, handling of the capsule by two workers, and taking ten more Rodac samples. Both workers rubbed their hands over all sampling surfaces and handed the capsule back and forth to each other several times. The capsule was not decontaminated again after the handling procedure.

The two investigators who occupied the downflow room for 2- and 4-hour periods were completely unrestricted in their movements. They wore only a surgical gown and performed such tasks as counting colonies, loading Andersen samplers, labeling plates, and wiping plates used in the Andersen samplers with isopropyl alcohol. They walked by the spacecraft and stainless steel table several times, leaned over them, and waved their arms over them. They extended their ungloved hands into the interior of the cone without touching it. All labor was performed within 3 or 4 ft of the spacecraft. The same tasks were performed in the crossflow room in addition to the labor that was performed by the 20 to 25 people who routinely worked in the room.

#### Statistical Analysis

The 95 percent upper and lower confidence limits for the reported values were computed and are indicated in Figures 4 through 13 by the shaded areas for each bar. These figures are merely graphical representations of the recorded values. For actual values, refer to the tables in the appendix. Where a statement was made that a difference was, or was not significant, a significance test was performed to test the null hypothesis that the reported value was the same under one condition or at one time as under another condition or time.

The confidence limits were constructed and the tests performed taking into account the fact that the reported values are coded functions of Poisson variables.

By coded is meant the reported value is the product of a constant and a count, which is assumed to follow the Poisson distribution. The estimated standard deviation of the reported value is the product of the constant and the square root of the count. The confidence limits on the reported values were developed by multiplying the limits of expectation appropriate to a count following the Poisson distribution by the constant. This procedure was followed for counts less than 50. For counts greater than 50, confidence limits were developed by treating the reported value as a normally distributed observation with standard deviations as previously described. Approximate significance tests of

differences between reported values were made by again treating the reported values as normal and applying the standard procedure for testing whether two normally distributed observations came from populations with the same mean.

When the airflow was off in the downflow room (Figure 4A), the variation among the eight counts obtained from the air was substantially in excess of that based on Poisson considerations. This indicated heterogeneity in the particle density throughout the room and from one day to another. Confidence limits in the mean number of viable particles per cubic foot were calculated in the usual fashion appropriate to a random sample from a normal population. (See appendix.)

## RESULTS

### Level of Microbial Contamination in the Air of the Laminar Downflow Room

Figure 4 shows the level of contamination in the air of the downflow room. Values plotted are the averages of counts obtained on 2 separate days for each time interval. The shaded areas indicate the 95 percent upper and lower confidence levels for the mean level of contamination. The viable particle count per cubic foot of air sampled with the Andersen sampler in the unoccupied room with the airflow off ranged from an average of 9.7 at zero sampling hr (15 minutes after decontamination) to 10.4 at 23 hr. The count dropped by more than half between the 0-hr and 2-hr samples and then climbed steadily for 23 hr. The air supply to the rooms had been turned off about 22 hr before taking the first sample at zero hr; therefore, the drop at 2 hr (24 hr after the air was turned off) may represent a die-off which was not noted at zero sampling hr (22 hr after air was turned off). The level of contamination built up again over the next 23 hr to approximately the same level as that measured at zero hr (Figure 4). If this presumption holds true, a 25-hr sample should be approximately half that of a 23-hr sample. This drop in the count at 2 hr was also noted when the airstream was on in both types of clean rooms (Figure 4 B, Figure 5), but because the counts were so low when the airstream was on the drop at 2 hr is probably insignificant.

The level of microbial contamination determined by the 0800 Monitor for airborne bacteria was considered unreliable because counts did not agree with other sampling procedures. For example, counts in the unoccupied downflow room with the airstream turned off were in the magnitude of 0.08 viable particles/ft<sup>3</sup> of air sampled (12 colonies total/drum) over a 23-hr period as compared to 9 to 10.4 viable particles/ft<sup>3</sup> of air sampled with the six-stage Andersen sampler (Figure 4A). When the airstream was turned on, no counts were obtained with the 0800 Monitor. The unusually low counts obtained with the 0800 Monitor were also noticed in the GSFC laboratory and the reason has not yet been resolved.

### Level of Microbial Contamination in the Air of the Laminar Crossflow Room

The level of contamination in the air of the occupied crossflow room was extremely low (0.5 viable particles/ft<sup>3</sup> or less), but was considerably higher than the counts obtained in the occupied downflow room. A comparison of Figure 4B and Figure 5 will show that, when the crossflow room was occupied for 0-, 2-, and 4-hr periods, the counts obtained downstream were 3 to 27 times greater than in the downflow room; the counts obtained upstream in the crossflow room were 3 to 18 times greater.

The same phenomenon noted in the downflow room occurred in the crossflow room (Figure 5); namely, counts were much lower at 2 hr than at 0 hr and increased steadily over the next 23 hr. Differences between counts obtained upstream and downstream of workers in the crossflow room were not statistically significant. A comparison of the data obtained from the air of the unoccupied crossflow and downflow rooms also showed that there were no statistically significant differences between the two rooms except at 0 hour.

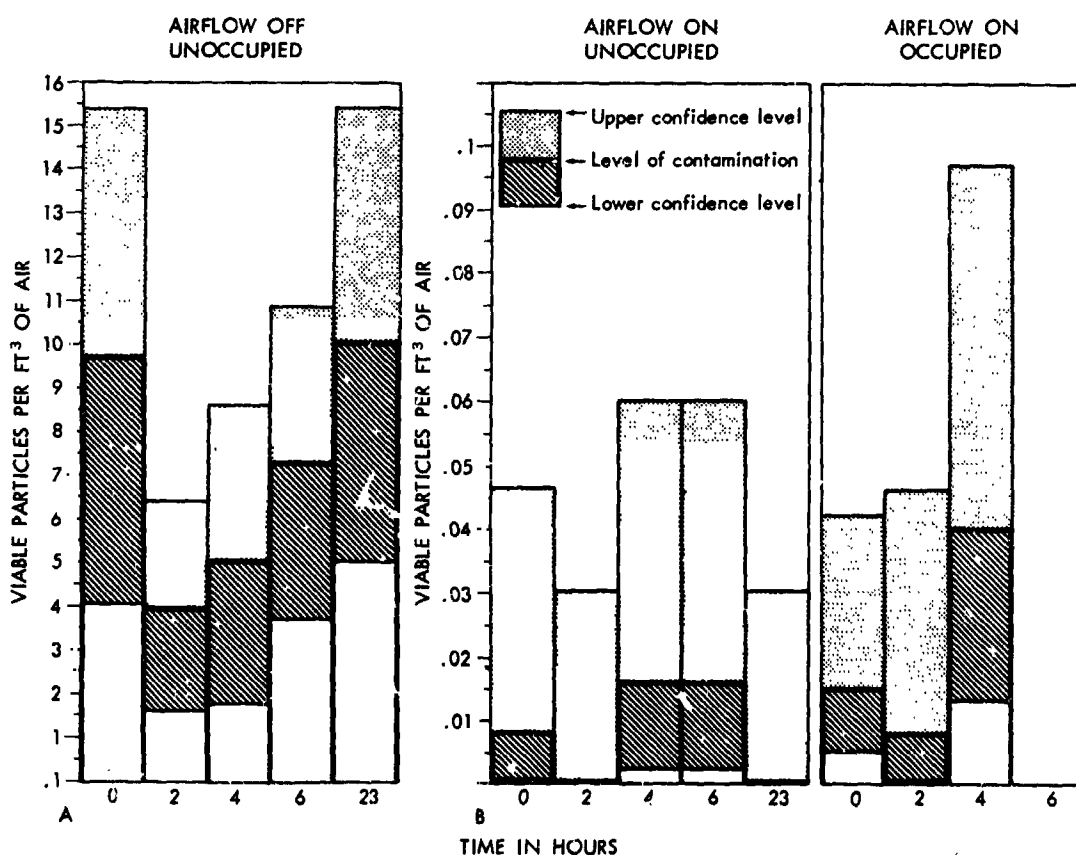


Figure 4. Level of Contamination of the Air in a Laminar Downflow Room

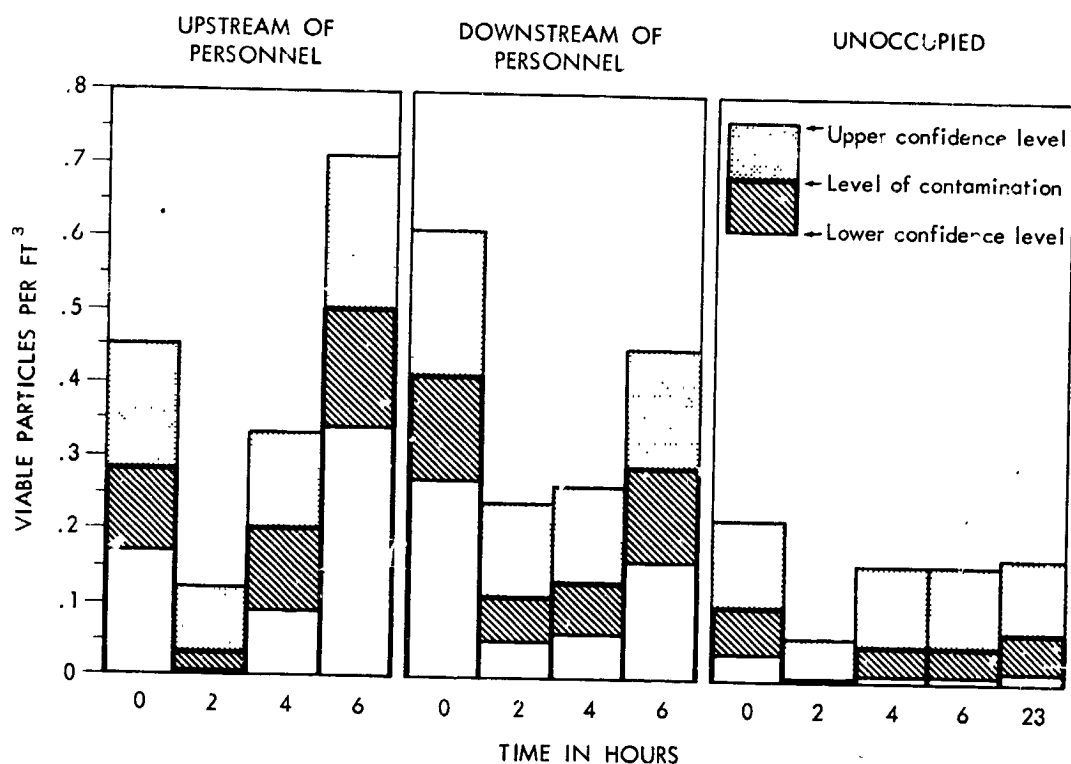


Figure 5. Level of Contamination of the Air in a Laminar Crossflow Room

Level of Microbial Contamination on the Surface of the Nosecone While Exposed in the Laminar Downflow Room

When the nosecone was exposed in the downflow room with the airflow turned off, counts were much higher on the interior surface than on the exterior surface (Figure 6). Values plotted in Figure 6 are the averages of counts obtained on 2 separate days on the entire exterior and interior surfaces. Counts on the interior surface ranged from 19 viable particles/ft² at 0 hr to 164 viable particles/ft² after 4 hr of exposure. On the exterior of the nosecone, counts ranged from 2.2 viable particles/ft² at 0 hour to 26 viable particles/ft² at 6 hr (Figure 6). On both the exterior and interior surfaces, the (viable) contamination increased for 4 and 6 hr respectively and then dropped off sharply over the 23-hr exposure period.

When the airstream was turned on, the level of contamination on the nosecone was greatly reduced, particularly on the interior surface (Figure 6). It can also be seen in Figure 6 that there were no statistically significant differences

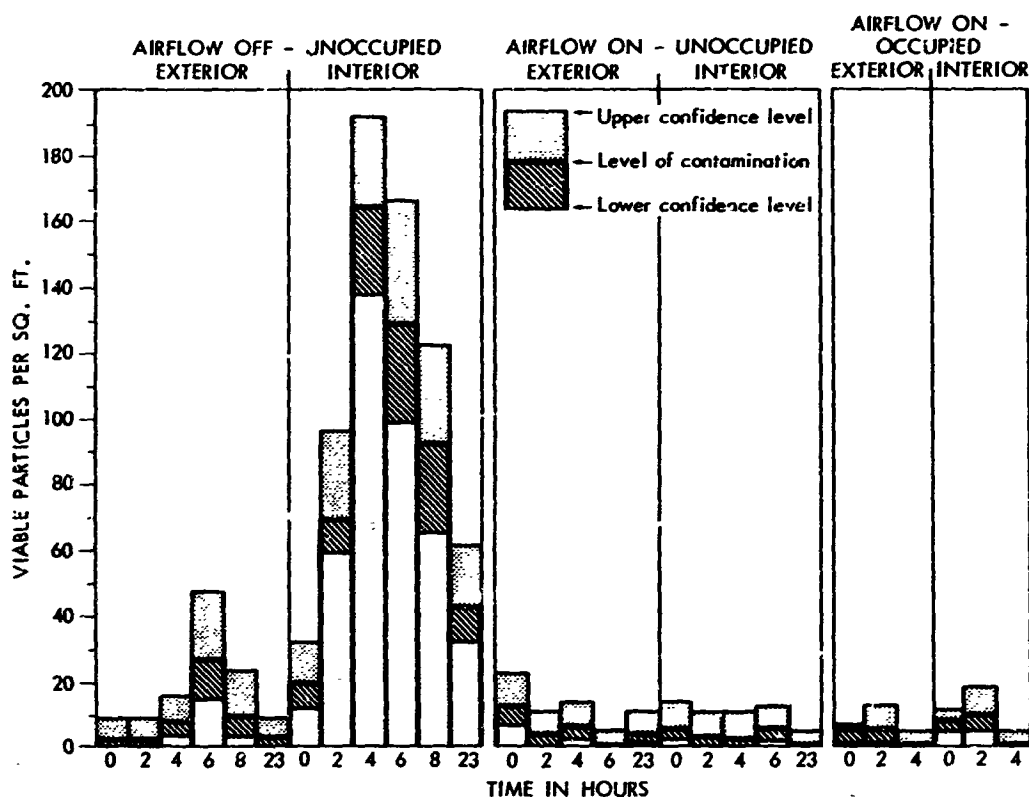


Figure 6. Level of Contamination on the Surface of a Nosecone While Exposed in a Laminar Downflow Room

in the level of contamination on the interior and exterior surfaces when the air was on. There was also no buildup of contamination on either surface over a 23-hr period as was noted when the airstream was off.

When the airstream in the downflow room was on, the presence of personnel for 2- and 4-hr periods appeared to have no significant effect on the level of contamination on the surface of the cone (Figure 6). The level of contamination was much the same as it was in the unoccupied room and even appeared to drop to zero after 4 hr. As indicated by the levels in Figure 6, the airstream exerts a cleaning effect on surfaces by keeping particulate matter (including microbes) suspended and moving away from the surface.

#### Level of Microbial Contamination on the Surface of the Nose Cone While Exposed in the Laminar Crossflow Room

Figure 7 shows the level of contamination on the interior and exterior surfaces of the nosecone while exposed in a laminar crossflow room as determined

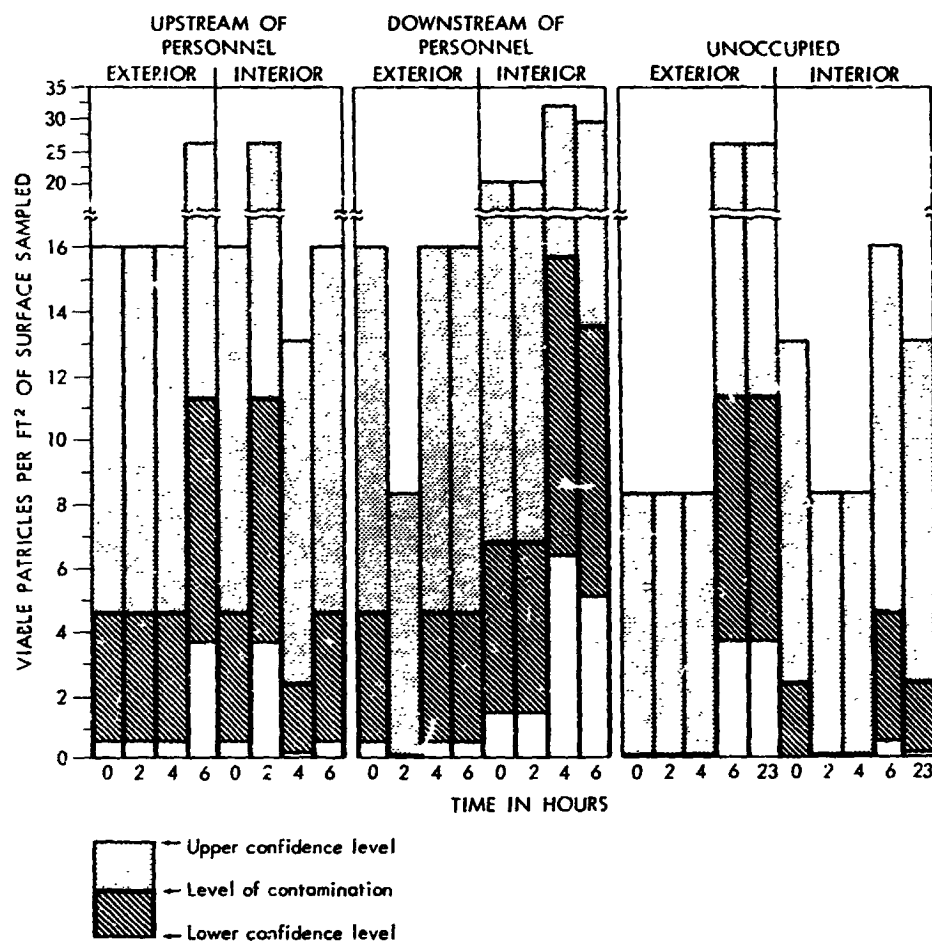


Figure 7. Level of Contamination on the Surface of a Nosecone While Exposed in a Laminar Crossflow Room

by the Rodac impression method. Values plotted under the three conditions (upstream, downstream, and unoccupied) are counts obtained on one workday for each condition, the unoccupied room being monitored on a Saturday. The airstream was turned on for all conditions.

The level of contamination was approximately the same on the exterior of the cone when exposed both upstream and downstream of the working personnel (Figure 7). However, the interior of the cone appeared to be slightly more contaminated when exposed downstream, particularly at 4 and 6 hr. Downstream, the level of contamination was greater on the interior than on the exterior surface of the cone, with peaks of 15.7 and 13.5 viable particles/ft<sup>2</sup> of surface sampled at 4 and 6 hr respectively; counts remained constant at 4.5 viable particles/ft<sup>2</sup> on the exterior except for the drop at 2 hr (Figure 7).

When the crossflow room was unoccupied, microbial contamination appeared to be reduced except on the exterior of the cone at 6 and 23 hr at which times it reached a peak of 11.2 viable particles/ft<sup>2</sup>, which is equivalent to the contamination count obtained in the occupied room (Figure 7, upstream). Although the scale is expanded in Figure 7, it should be noted that the level of contamination is very similar to that obtained in the downflow room with the airflow on.

#### Level of Microbial Contamination on Surface of the Mars Landing Capsule While Exposed in a Laminar Downflow Room

With the airstream turned off, microbial contamination accumulated on the surface of the landing capsule over a 23-hr period in the unoccupied downflow room. The count approximately doubled, rising from an average of 103 viable particles/ft<sup>2</sup> at 0 hr to 202 viable particles/ft<sup>2</sup> at 23 hr (Figure 8). The values plotted in Figure 8 are the averages of counts obtained on 2 separate days.

The cleaning effect of the airstream is seen again in Figure 8, when the airstream is turned on. Microbial contamination was greatly reduced and no contamination accumulated over a 23-hr period; the count dropped from 7.2 viable

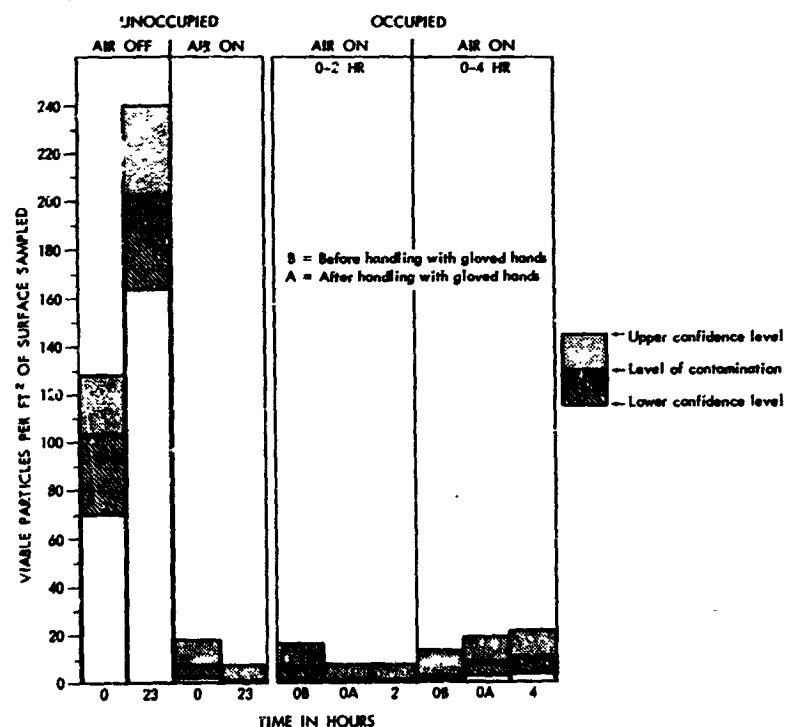


Figure 8. Level of Contamination on the Surface of a Landing Capsule While Exposed in a Laminar Downflow Room

particles/ft<sup>2</sup> at 0 hr to 0 viable particles/ft<sup>2</sup> at 23 hr. The counts obtained were reproducible, as the 0 counts in Figure 8 illustrate (average of 2 days).

The introduction of two people into the downflow room for 2- and 4-hr periods with the airstream turned on did not significantly add to the microbial contamination on the surface of the capsule. Figure 8 shows that microbial contamination was actually higher at 0 hr (6 viable particles/ft<sup>2</sup>) than it was after 2 hr of occupation (0 count). The microbial contamination on the surface of the capsule increased slightly after 4 hr of occupation, rising from 2 viable particles/ft<sup>2</sup> at 0 hr to 10 viable particles/ft<sup>2</sup> after 4 hr of occupation. It is interesting to note that handling of the capsule by two workers with gloved hands did not greatly increase the microbial contamination (Figure 8); in fact, counts were usually lower after the capsule was handled than before the capsule was handled or remained the same (see crossflow room, Figure 9).

#### Level of Microbial Contamination on the Surface of the Mars Landing Capsule While Exposed in a Laminar Crossflow Room

Figure 9 shows the level of microbial contamination on the surface of the Mars landing capsule exposed in a laminar crossflow room. The airstream was on at all times and the values plotted are counts obtained on a single day for each sampling condition (upstream, downstream, and unoccupied). One observes first in Figure 9 the significantly higher level of microbial contamination on the surface of the capsule exposed downstream of the 20 to 25 working personnel. The contamination rose steadily from 14.4 particles/ft<sup>2</sup> at 0 hr to 58.0 viable particles/ft<sup>2</sup> at 6 hr. Counts were much lower when the capsule was exposed upstream of personnel, and the contamination did not build up as it did downstream.

The results obtained when the capsule was exposed in the unoccupied crossflow room closely paralleled those obtained when the capsule was exposed upstream and were actually slightly higher, but perhaps not significantly so (Figure 9). In the unoccupied room, counts obtained from the capsule decreased from 14.4 viable particles/ft<sup>2</sup> at 0 hr to 3.6 viable particles/ft<sup>2</sup> at 23 hr (a 75 percent reduction); when exposed upstream of working personnel, counts decreased from 11 viable particles/ft<sup>2</sup> at 0 hr to 3.6 viable particles/ft<sup>2</sup> at 6 hr (a 68 percent reduction).

As noted in the downflow room (Figure 8), counts obtained from the capsule after being handled with gloved hands were lower than, or the same as, before being handled (Figure 9). This result is remarkable, considering that Rodac samples were taken immediately after handling the capsule and that sampling before and after handling was accomplished in less than 10 min. Note that the two investigators who took the samples used the same gloved hands to handle



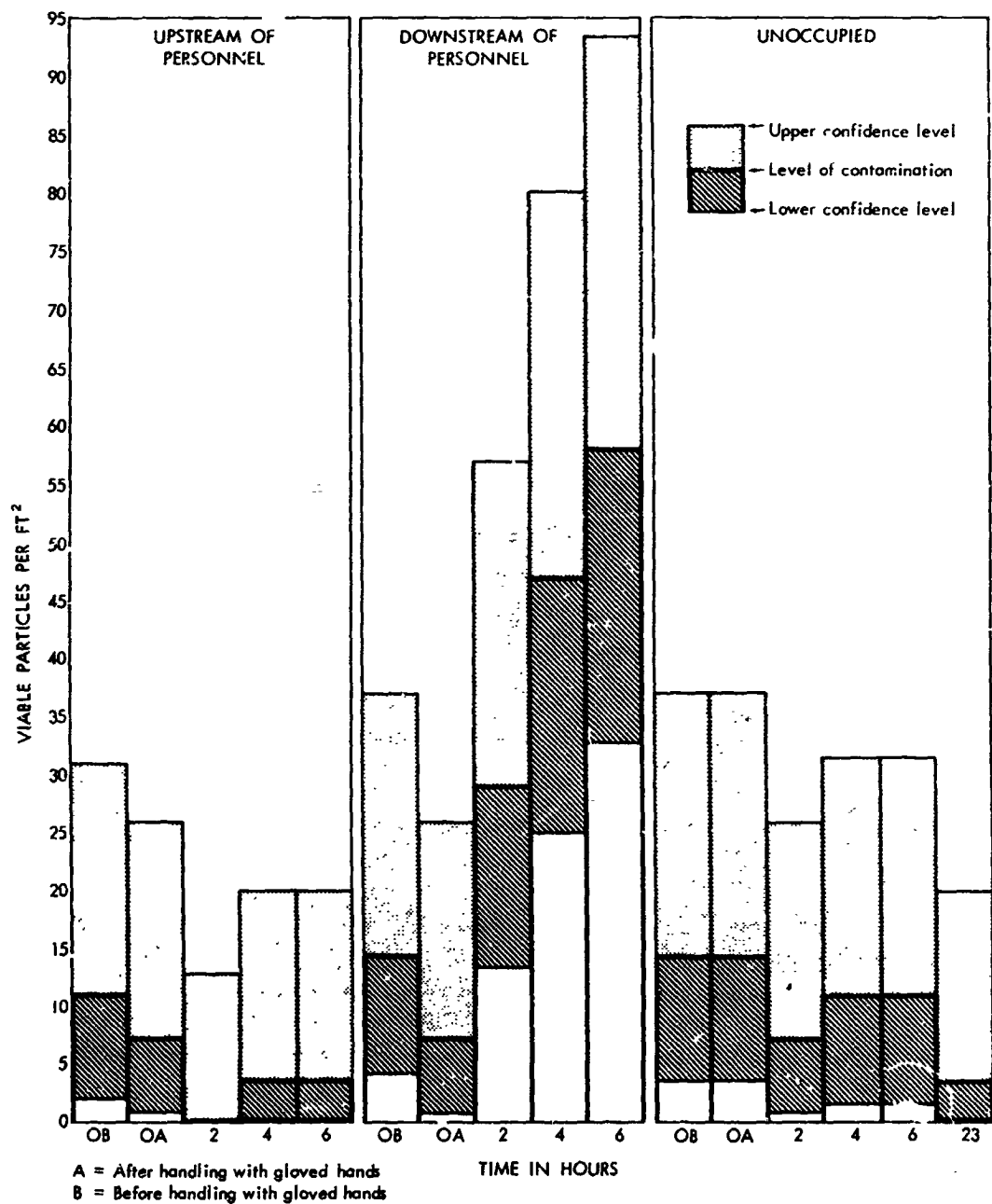


Figure 9. Level of Contamination on the Surface of a Landing Capsule While Exposed in a Laminar Crossflow Room

Rodac plates and to sample the nosecone before sampling the capsule. Yet, by observing proper technique and by keeping their hands away from face, body, and extraneous objects, handling of the capsule with gloved hands did not increase the microbial load.

### Level of Microbial Contamination on the Surface of a Stainless Steel Table Exposed in a Laminar Downflow Room

Figure 10 shows the level of microbial contamination on the surface of a stainless steel table exposed in a laminar downflow room. Values plotted are

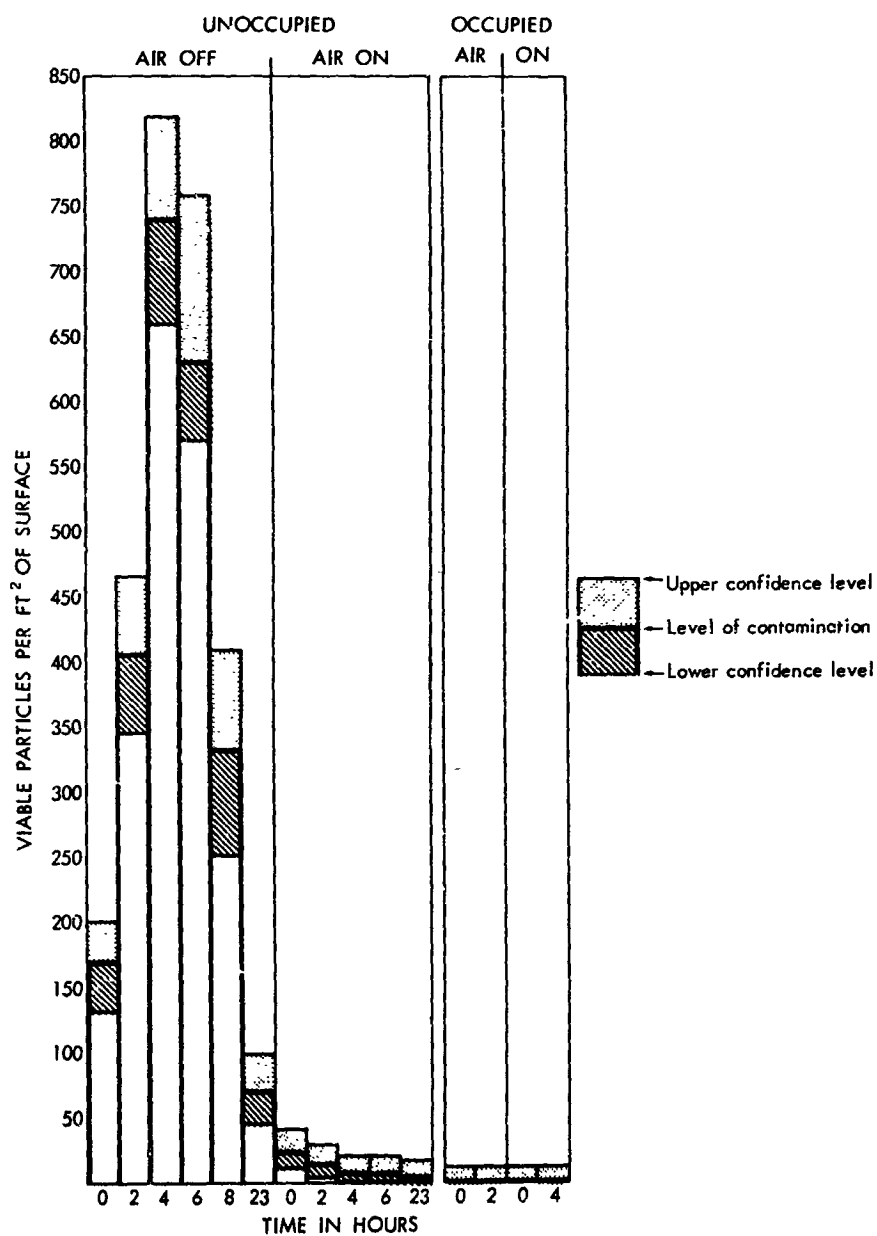


Figure 10. Level of Contamination on the Surface of a Stainless Steel Table While Exposed in a Laminar Downflow Room

averages of 2 determinations. When the airstream was turned off in the unoccupied room, the contamination on the table increased from 171 viable particles/ft<sup>2</sup> at 0 hr to 740 hr at 4 hr (a fourfold increase). The count decreased quite rapidly after 4 hr to 70 viable particles/ft<sup>2</sup> at 23 hr (a decrease of 60 percent and 91 percent from 0 and 4 hr respectively).

By turning the airstream on in the downflow room, the contamination was reduced by 87 percent at 0 hr, by 99 percent at 4 hr, and by 92 percent at 23 hr (Figure 10). It can also be seen in Figure 10 that the microbial contamination decreased progressively from 23 viable particles/ft<sup>2</sup> at 0 hr to 5 viable particles/ft<sup>2</sup> at 23 hr when the airstream was on in the unoccupied downflow room.

Occupation of the downflow room with the air on did not result in any contamination after 0, 2, and 4 hr (Figure 10). The results obtained with the stainless steel table agree with results obtained with the landing capsule; i.e., personnel inhabiting the laminar downflow room did not seem to contribute to the level of microbial contamination on surfaces or in the air. The variable counts obtained at 0 hr probably reflect the level of decontamination exercised. Although attempts were made to standardize the decontamination procedure, it appeared to be more effective at some times than at others.

#### Level of Microbial Contamination on the Surface of a Stainless Steel Table While Exposed in a Laminar Crossflow Room

Figure 11 shows the microbial contamination on a stainless steel table exposed in a laminar crossflow room. Values plotted are counts obtained on a single day for each condition (upstream, downstream, and unoccupied). The contamination was much greater on the table top during exposure downstream of working personnel than during exposure either upstream or in the unoccupied room. As noted on the capsule (Figure 9) and on the interior of the nosecone (Figure 7) the contamination appeared to build up when the table was downstream of personnel. Counts increased from 9 viable particles/ft<sup>2</sup> at 0 hr to 40 viable particles/ft<sup>2</sup> at 6 hr, a fourfold increase. This buildup of contamination was not observed upstream of personnel.

The microbial contamination at 0 hr was the same upstream as downstream (9 viable particles/ft<sup>2</sup>). However, the count upstream decreased to 0 at 2 and 4 hr and then increased slightly to 4.5 viable particles/ft<sup>2</sup> at 6 hr, which still represented a decrease of 50 percent from 0 hr. It should be borne in mind that extremely low levels of contamination were being measured when the airstream was on, not only on the table top, but throughout this study, and that 4.5 viable particles/ft<sup>2</sup> represents a total of only 1 colony per 8 Rodac plates used for sampling the table.

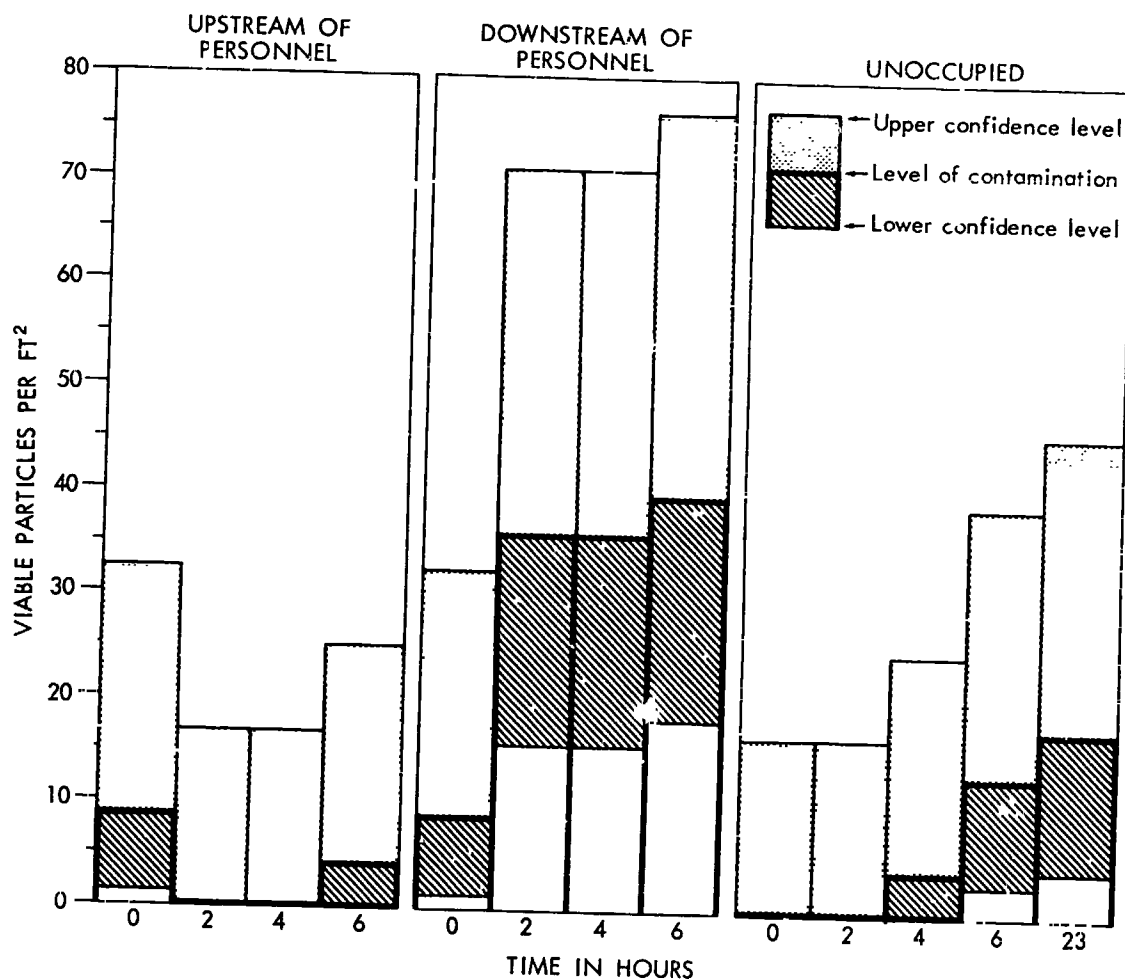


Figure 11. Level of Contamination on the Surface of a Stainless Steel Table While Exposed in a Laminar Crossflow Room

When the table top was monitored in the unoccupied crossflow room, the counts were zero at 0 and 2 hr and then increased from 4.5 viable particles/ft<sup>2</sup> at 4 hr to 18 viable particles/ft<sup>2</sup> at 23 hr (a fourfold increase). Again, the contamination on the table in the unoccupied crossflow room was not as great as it appears to be in Figure 11 when one realizes that the counts increased from 1 colony per 8 Rodac plates at 4 hr (4.5 viable particles/ft<sup>2</sup>) to only 4 colonies per 8 Rodac plates (18 viable particles/ft<sup>2</sup>) at 23 hr.

#### Microbial Fallout in a Laminar Downflow Room

Figure 12 shows microbial fallout in a laminar downflow room. Values plotted are the averages of counts obtained on 2 separate days. Counts were

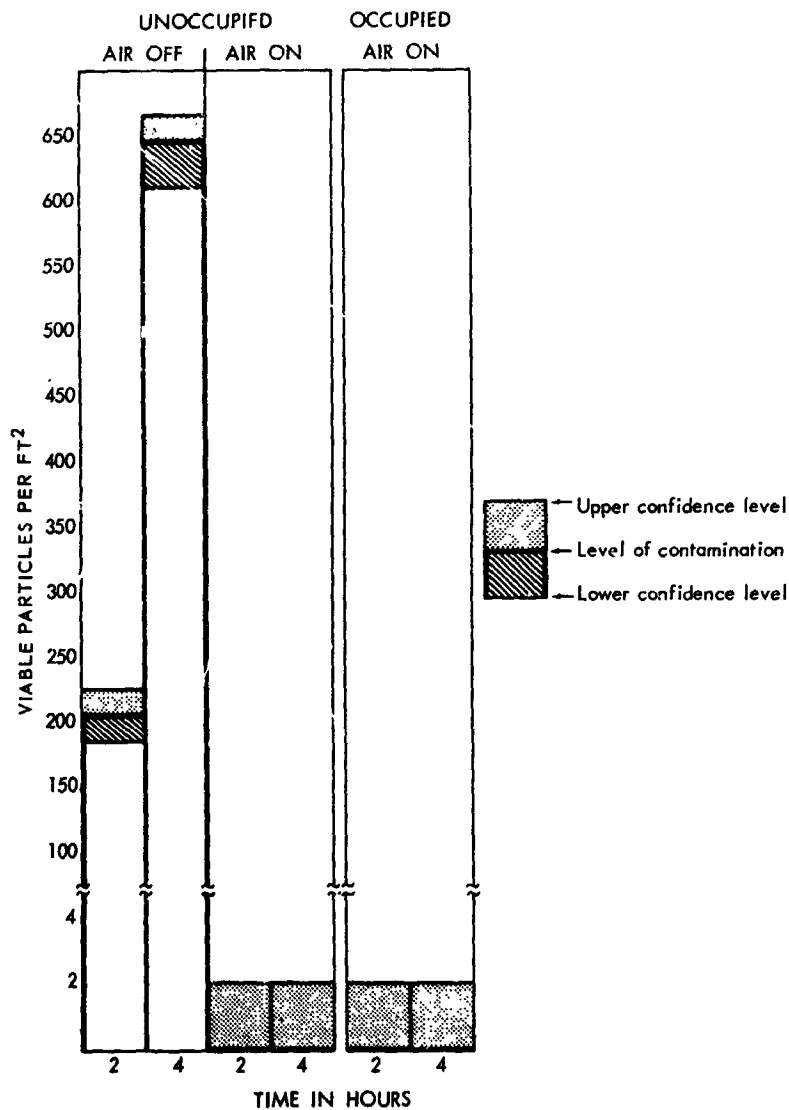


Figure 12. Microbial Fallout on Blood Agar Plates in a Laminar Downflow Room

quite high in the unoccupied room when the airstream was turned off, ranging from 204 viable particles/ft<sup>2</sup> after 2 hr of exposure to 646 viable particles/ft<sup>2</sup> after 4 hr exposure (a threefold increase).

When the airstream was turned on, counts were zero after 2- and 4-hr exposures, even when the room was occupied. Although not shown in Figure 12, counts in the three areas of the room were very similar.

### Microbial Fallout in a Laminar Crossflow Room

Microbial contamination on blood agar settling plates was 12 to 20 times greater downstream of working personnel than it was either upstream or in the unoccupied room (Figure 13). Values plotted in Figure 13 are counts obtained on a single day for each sampling condition.

Counts were approximately the same after 2- and 4-hr exposures upstream (5 and 3 viable particles/ft<sup>2</sup> respectively). In the unoccupied room, counts were zero after 2-hr exposure and rose to 6 viable particles/ft<sup>2</sup> after 4-hr exposure. More data are needed to make definite conclusions, but there is some indication that contamination was approximately the same upstream and in the unoccupied room and was much greater downstream as determined by fallout plates. There also appeared to be more microbial fallout when the airstream was on in the crossflow room (Figure 13) than in the downflow room (Figure 12).

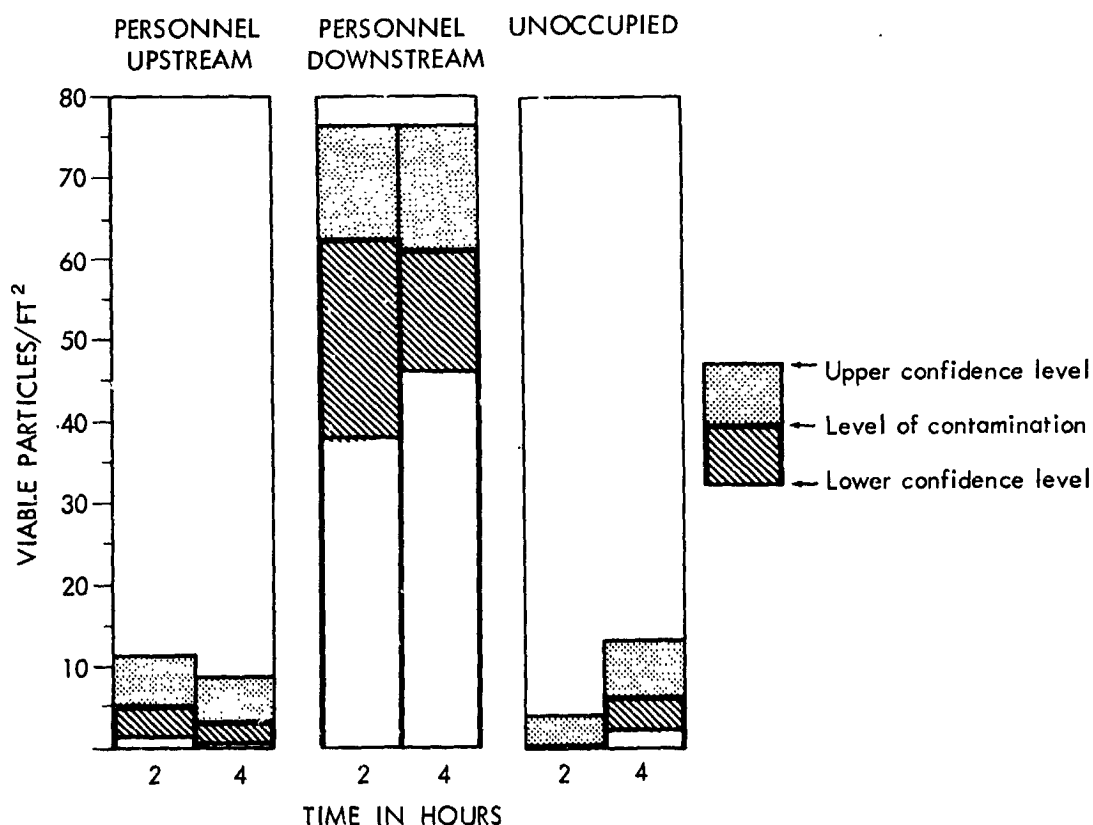


Figure 13. Microbial Fallout on Blood Agar Plates in a Laminar Crossflow Room

## DISCUSSION

The concentration of viable particles detected in the air and on surfaces in both types of laminar flow clean rooms was far less than maximum allowable levels specified by NASA. In the "Interim Requirements for Bioclean Facilities for Planetary Spacecraft" (ref. 13), NASA requires that bioclean room air not exceed an average of 2 viable particles/ft<sup>3</sup> of air for any 10 successive samples. This study demonstrated that the highest concentration of viable particles in the occupied downflow room over a 4-hr period (Figure 4B) did not exceed an average of 0.04 viable particles/ft<sup>3</sup> of air sampled, 98 percent less than required by the NASA standard. The air of the crossflow room during occupation by 20 to 25 personnel, who assembled electronic circuits, had a maximum concentration of viable particles, 75 percent less than required by the NASA standard over a period of 6 hr (Figure 5). A comparison of the two clean rooms is valid because the number of people per square foot of space was greater in the downflow room by at ratio of 2:1.

The degree of microbial contamination on the surface of the cone, capsule, and table top while exposed in both the downflow and crossflow rooms was far below the 200 viable particles/ft<sup>2</sup> of surface required by NASA for bioclean rooms, as determined by the Rodac impression technique. The counts were higher on the three kinds of surfaces when they were exposed downstream of personnel than when they were upstream of personnel in the crossflow room, but still remained far below 200 viable particles/ft<sup>2</sup> of surface.

Counts obtained from the air of the occupied downflow room were 3 to 27 times less than counts obtained upstream or downstream in the occupied crossflow room over a 4-hr period when the airstream was turned on. When the rooms were unoccupied, however, there appeared to be little difference in the air of the two rooms. Personnel in the downflow room did not greatly increase the degree of microbiological contamination in the air, but in the crossflow room counts in the air were higher both upstream and downstream of personnel than in the unoccupied room.

The contamination on the surface of the nosecone, capsule, and steel table top did not increase significantly when personnel were introduced into the downflow room for 4 hr, but counts on these same surfaces in the crossflow room were increased by the presence of personnel upstream of them.

As mentioned in the results, handling of the capsule with gloved hands by two investigators did not appear to increase significantly the microbiological contamination of its surface in either clean room. This result indicates that employing sterile gloves as well as sterile clothing will considerably minimize microbial loading by personnel. Employment of proper techniques can also minimize the contamination, both in the air and on surfaces.

The airstream is extremely efficient in removing viable airborne contaminants from the clean room, as was demonstrated dramatically in this study when the airstream was turned on. The airstream, particularly in the downflow room, not only reduced counts in the air, but also on surfaces. It was also noticed in the downflow room that contamination did not build up on surfaces when the airstream was on. The efficiency of this same downflow room in removing viable particles was also demonstrated by Peakly (ref. 14), who aerosolized Serratia marcescens into the room at a concentration of  $5 \times 10^5$  cells/ft<sup>3</sup> of air in the room. Following 1 minute of operation, no viable particles of Serratia marcescens were detected by Andersen samplers.

More work is needed to evaluate these clean rooms. However, this preliminary study indicates that the microbiological contamination of air and surfaces can be kept at a lower level in a laminar downflow room than in a laminar cross-flow room and, conceivably, than in a so-called conventional clean room (no laminar airflow). This study also indicates that personnel working in the downflow room will be less restricted and will have considerably more freedom to move around than in other types of clean rooms; the laminar airstream makes this possible by removing the contamination from the room as it is shed by personnel. Working over the capsule, cone, and table did not increase the microbial contamination in the downflow room, even though the investigators leaned over them with uncovered head, face, and hands.



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## APPENDIX

### Mathematical Calculations

1. The total number of viable particles per square foot of surface area sampled was determined by the following calculation:

$$\text{Total microorganisms per ft}^2 = \frac{\text{Total count} \times 144 \text{ sq. in.}}{4.0 \times \# \text{ plates}}$$

where 4.0 = surface area of Rodac plate in sq. in.

2. The number of viable particles settling on a surface (blood agar plates) was determined by the same formula as shown above, except that 9.6 sq. in. was the surface area of the petri plate.
3. The number of viable particles per cubic foot of air sampled by the Andersen sampler was determined by the following calculation:

$$\frac{\text{Total Count}}{4 \text{ Andersen samplers} \times 2 \text{ days} \times 15 \text{ min}}$$

Example 1: Air sampling, downflow, airflow off, unoccupied, 0 hours

Total count for 4 Andersen samples = 1165

$$\therefore \frac{1165}{4 \times 2 \times 15} = \frac{1165}{120} = 9.7$$

4. The confidence level was calculated as follows for counts greater than 50:

R = Mean value reported in the figures

D = 4 Andersen samplers  $\times$  2 days  $\times$  15 minutes = 120

T = Total count

T/D = R

L = Lower confidence limit =  $(1/D) (T - 1.96 \sqrt{T})$

U = Upper confidence limit =  $(1/D) (T + 1.96 \sqrt{T})$

5. Where total counts going into the reported figure were less than 50, the 95 percent confidence limits were computed as follows:

- a. Find total ( T ) of all counts going into the reported figure.
- b. Find the divisor ( D ) of the total count which yields the reported figure.  
The divisor for various cases were determined as follows:

| <u>Case</u>     | <u>Divisor</u>  |
|-----------------|---|
| Air sampling    | $15 \times$ number of independent Andersen samples going into total |
| Rodac sampling  | $(4/144) \times$ number of Rodac plates going into total            |
| Settling plates | $(9.6/144) \times$ number of agar plates going into total           |

c. Read off TL (lower limit of expectation) and TU (upper limit of expectation) from a table by Pearson and Hartley (ref. 15).

d. Compute the 95 percent confidence limits as:

$$L = TL/D \text{ and } U = TU/D$$

Example 2: Sampling of exterior cone, downflow room, airflow off, unoccupied, 0 hour.

$$T = \text{Total count} = 2$$

$$D = .88889 = \frac{4 \text{ sq. inches} \times 16 \text{ Rodac plates} \times 2 \text{ days}}{144 \text{ sq. inches (1 ft}^2\text{)}}$$

$$TL = .242$$

$$TU = 7.22$$

$$L = TL/D = .242/.88889 = .207$$

$$U = TU/D = 7.22/.88889 = 8.1$$

6. The data from the air of the unoccupied downflow room, with the airflow turned off (Figure 4A), did not follow a Poisson distribution, so calculations in this case only were based on a normal population. Using the eight observations (mean count for each Andersen sampler), a sample variance, a standard error of the sample mean, and the upper and lower 95 percent confidence

limits were computed in the usual manner appropriate to a random sample of observations from a normal population. The formulas employed were:

a.  $S^2 = \frac{\sum(x - \bar{x})^2}{N - 1}$  = Sample variance among observations

b.  $\frac{S\bar{x} - S}{\sqrt{N}}$  = Standard error of sample mean

c.  $LL = \bar{x} - ts\bar{x}$  = Lower confidence level

d.  $UL = \bar{x} + ts\bar{x}$  = Upper confidence level

7. The significance test, to test whether reported value  $R_1 = T_1/D_1$  is different from reported value  $R_2 = T_2/D_2$ , was calculated as follows:

$$T_0 = \frac{R_1 - R_2}{\sqrt{T_1/D_1^2 + T_2/D_2^2}}$$

Example 3: Air sampling, downflow, airflow off, unoccupied. To test the null hypothesis that the reported value at 0 hours is the same as at 2 hours and having the following data (see Figure 4A):

$$\begin{array}{ll} R_1 = 9.7 & R_2 = 4.0 \\ T_1 = 1165 & T_2 = 485 \\ D_1 = 120 & D_2 = 120 \end{array}$$

Thus:

$$T_0 = \frac{R_1 - R_2}{\sqrt{T_1/D_1^2 + T_2/D_2^2}} = \frac{9.7 - 4.0}{\sqrt{1165/(120)^2 + 485/(120)^2}} = 16$$

Since  $|T_0| > 1.96$ , there is evidence that the reported values are different. The test is a two tailed test with a significance level of 5 percent.

**TABLE A-1**  
**Level of Contamination of the Air in a Laminar Downflow Room**  
(values in viable particles per cubic foot)

| Sampling Time<br>(in hours) | AIRFLOW OFF - UNOCCUPIED                    |                              |                              | AIRFLOW ON - UNOCCUPIED                     |                              |                              | AIRFLOW ON - OCCUPIED                       |                              |   |                              |                              |
|-----------------------------|---|------------------------------|------------------------------|---|------------------------------|------------------------------|---|------------------------------|---|------------------------------|------------------------------|
|                             | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |
| 0                           | 9.7   | 15.42                        | 4.06                         | 0.008                                       | 0.048                        | 0.0002                       | 0.015                                       | 0.042                        | 0.015                                       | 0.042                        | 0.005                        |
| 2                           | 4.0   | 6.43                         | 1.65                         | 0   | .03                          | 0                            | .008  | .046                         | .008  | .046                         | .0002                        |
| 4                           | 5.1   | 8.61                         | 1.71                         | .016  | .06                          | .002                         | .04   | .097                         | .04   | .097                         | .013                         |
| 6                           | 7.3   | 10.89                        | 3.71                         | .016  | .06                          | .002                         |   |                              |   |                              |                              |
| 23                          | 10.4  | 15.46                        | 5.04                         | 0   | .03                          | 0                            |   |                              |   |                              |                              |

Note: Table corresponds to Figure 4.

TABLE A-2  
Level of Contamination of the Air in a Laminar Crossflow Room  
(values in viable particles per cubic foot)

| Sampling Time<br>(in hours) | UPSTREAM OF PERSONNEL                       |                              |                              |   | DOWNSTREAM OF PERSONNEL      |                              |   |                              | UNOCCUPIED                   |   |                              |                              |
|-----------------------------|---|------------------------------|------------------------------|---|------------------------------|------------------------------|---|------------------------------|------------------------------|---|------------------------------|------------------------------|
|                             | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level | Viable<br>Particles<br>per ft. <sup>3</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |
| 0                           | 0.28  | 0.45                         | 0.17                         | 0.41  | 0.61                         | 0.27                         | 0.1   | 0.22                         | 0.037                        |   |                              |                              |
| 2                           | .03   | .12                          | .004                         | .11   | .24                          | .05                          | 0   | .062                         | 0                            |   |                              |                              |
| 4                           | .2  | .33                          | .09                          | .13   | .26                          | .06                          | .05   | .16                          | .01                          |   |                              |                              |
| 6                           | .5  | .71                          | .34                          | .29   | .45                          | .16                          | .05   | .16                          | .01                          |   |                              |                              |
| 23                          |   |                              |                              |   |                              |                              | .07   | .171                         | .016                         |   |                              |                              |

Note: Table corresponds to Figure 5.

TABLE A-3

Level of Contamination on the Surface of a Nosecone While Exposed in a Laminar Downflow Room  
(values in viable particles per square foot)

| Sampling Time (in hours) | AIRFLOW OFF - UNOCCUPIED              |                           |                           |                                       |                           |                           | AIRFLOW ON - UNOCCUPIED               |                           |                           |                                       |                           |                           | AIRFLOW ON - OCCUPIED                 |                           |                           |                                       |                           |                           |
|--------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|
|                          | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           |
|                          | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level |
| 0                        | 2                                     | 8                         | 0.207                     | 19                                    | 31                        | 11                        | 12                                    | 22                        | 6                         | 5.6                                   | 13                        | 2                         | 4.6                                   | 6                         | 0.9                       | 7                                     | 11                        | 4                         |
| 2                        | 2                                     | 8                         | 0.207                     | 79                                    | 96                        | 59                        | 3                                     | 10                        | 0.72                      | 3                                     | 10                        | 0.7                       | 4.5                                   | 12                        | 1                         | 9                                     | 18                        | 4                         |
| 4                        | 7                                     | 15                        | 2.5                       | 164                                   | 191                       | 137                       | 5.5                                   | 13                        | 2                         | 2                                     | 10                        | 0.7                       | 0                                     | 4                         | 0                         | 0                                     | 4                         | 0                         |
| 6                        | 4.3                                   | 47                        | 14                        | 128                                   | 166                       | 90                        | 0                                     | 4                         | 0                         | 4.5                                   | 12                        | 1                         |                                       |                           |                           |                                       |                           |                           |
| 8                        | 9                                     | 23                        | 2.4                       | 92                                    | 122                       | 65                        |                                       |                           |                           |                                       |                           |                           |                                       |                           |                           |                                       |                           |                           |
| 23                       | 2.4                                   | 9                         | 0.207                     | 42                                    | 61                        | 32                        | 3                                     | 10                        | 0.7                       | 0                                     | 4                         | 0                         |                                       |                           |                           |                                       |                           |                           |

Note: Table corresponds to Figure 6.

TABLE A-4

Level of Contamination on the Surface of a Nosecone While Exposed in a Laminar Crossflow Room  
(values in viable particles per square foot)

| Sampling Time (in hours) | UPSTREAM OF PERSONNEL                 |                           |                           |                                       |                           |                           | DOWNSTREAM OF PERSONNEL               |                           |                           |                                       |                           |                           | UNOCCUPIED                            |                           |                           |                                       |                           |                           |
|--------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|---------------------------------------|---------------------------|---------------------------|
|                          | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           | EXTERIOR                              |                           |                           | INTERIOR                              |                           |                           |
|                          | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level | Viable Particles per ft. <sup>2</sup> | Upper Confid- dence Level | Lower Confid- dence Level |
| 0                        | 4.5                                   | 16                        | 0.5                       | 4.5                                   | 16                        | 0.5                       | 4.5                                   | 16                        | 0.5                       | 6.7                                   | 20                        | 1.1                       | 0                                     | 8.3                       | 0                         | 2.2                                   | 13                        | 0.06                      |
| 2                        | 4.5                                   | 16                        | 0.5                       | 11.2                                  | 26                        | 3.6                       | 0                                     | 8.3                       | 0                         | 6.7                                   | 20                        |                           | 0                                     | 8.3                       | 0                         | 0                                     | 8.3                       | 0                         |
| 4                        | 4.5                                   | 16                        | 0.5                       | 2.2                                   | 13                        | 0.06                      | 4.5                                   | 16                        | 0.5                       | 15.7                                  | 32                        |                           | 0                                     | 8.3                       |                           | 0                                     | 8.3                       | 0                         |
| 6                        | 11.2                                  | 26                        | 3.6                       | 4.5                                   | 16                        | 0.5                       | 4.5                                   | 16                        | 0.5                       | 13.5                                  | 29.5                      | 5                         | 11.2                                  | 26                        | 3.6                       | 5                                     | 16                        | 0.5                       |
| 23                       |                                       |                           |                           |                                       |                           |                           |                                       |                           |                           |                                       |                           |                           | 11.2                                  | 26                        | 3.6                       | 2.2                                   | 13                        | 0.06                      |

Note: Table corresponds to Figure 7.

TABLE A-5

Level of Contamination on the Surface of a Landing Capsule While Exposed in a Laminar Downflow Room

(values in viable particles per square foot)

| Sampling Time<br>(in hours) | UNOCCUPIED                                  |                                   |                                   |   |                                   | OCCUPIED                          |   |                                   |   |                                   |
|-----------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|---|-----------------------------------|
|                             | AIR OFF                                     |                                   | AIR ON                            |   |                                   | AIR ON, 0-2 HR.                   |   |                                   | AIR ON, 0-4 HR.                             |                                   |
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Lower<br>Confi-<br>dence<br>Level |
| 0                           | 103   | 128                               | 70                                | 7.2   | 18                                | 2                                 |   |                                   |   |                                   |
| 23                          | 202   | 240                               | 164                               | 0   | 6.6                               | 0                                 |   |                                   |   |                                   |
| 0B                          |   |                                   |                                   |   |                                   |                                   | 6   | 16                                | 2   | 13                                |
| 0A                          |   |                                   |                                   |   |                                   |                                   | 0   | 7                                 | 8   | 19                                |
| 2                           |   |                                   |                                   |   |                                   |                                   | 0   | 7                                 |   |                                   |
| 4                           |   |                                   |                                   |   |                                   |                                   |   |                                   | 10  | 21                                |
|                             |   |                                   |                                   |   |                                   |                                   |   |                                   |   | 3                                 |

Note: (1) Table corresponds to Figure 8.

(2) A = After handling with gloved hands.

(3) B = Before handling with gloved hands.



TABLE A-6  
Level of Contamination on the Surface of a Landing Capsule While Exposed in a Laminar Crossflow Room  
(values in viable particles per square foot)

| Sampling Time<br>(in hours) | UPSTREAM OF PERSONNEL                       |                              |                              |  | DOWNSTREAM OF PERSONNEL                     |                              |                              |  | UNOCCUPIED                                  |                              |                              |  |
|-----------------------------|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|--|
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  |
| 0B                          | 11  | 31                           | 2                            |  | 14.4  | 37                           | 3.94                         |  | 14.4  | 37                           | 3.6                          |  |
| 0A                          | 7.2   | 26                           | 0.9                          |  | 7.2   | 26                           | 0.9                          |  | 14.4  | 37                           | 3.6                          |  |
| 2                           | 0   | 12.8                         | 0                            |  | 29  | 57                           | 13.5                         |  | 7.2   | 26                           | 0.9                          |  |
| 4                           | 3.6   | 20                           | 0.091                        |  | 47  | 80                           | 25                           |  | 11  | 31.6                         | 2                            |  |
| 6                           | 3.6   | 20                           | 0.091                        |  | 58  | 93.5                         | 33                           |  | 11  | 31.6                         | 2                            |  |
| 23                          |   |                              |                              |  |   |                              |                              |  | 3.6   | 20                           | .091                         |  |

Note: (1) Table corresponds to Figure 9.  
(2) A = After handling with gloved hands.  
(3) B = Before handling with gloved hands.

TABLE A-7  
Level of Contamination on the Surface of a Stainless Steel Table While Exposed in a Laminar Downflow Room  
(values in viable particles per square foot)

| Sampling Time<br>(in hours) | UNOCCUPIED                                  |                                   |                                   |   |                                   |                                   | OCCUPIED                                    |                                   |                                   |   |                                   |                                   |
|-----------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|
|                             | AIR OFF                                     |                                   |                                   | AIR ON                                      |                                   |                                   | AIR ON, 0-2 HR                              |                                   |                                   | AIR ON, 0-4 HR                              |                                   |                                   |
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level |
| 0                           | 171   | 201                               | 132                               | 23  | 41                                | 11                                | 0   | 8                                 | 0                                 | 0   | 8                                 | 0                                 |
| 2                           | 404   | 465                               | 346                               | 14  | 29                                | 5                                 | 0   | 8                                 | 0                                 |   |                                   |                                   |
| 4                           | 740   | 820                               | 660                               | 7   | 20                                | 1                                 |   |                                   |                                   | 0   | 8                                 | 0                                 |
| 6                           | 630   | 760                               | 570                               | 7   | 20                                | 1                                 |   |                                   |                                   |   |                                   |                                   |
| 8                           | 333   | 410                               | 252                               |   |                                   |                                   |   |                                   |                                   |   |                                   |                                   |
| 23                          | 70  | 97                                | 46                                | 5   | 16                                | 0.5                               |   |                                   |                                   |   |                                   |                                   |

Note: Table corresponds to Figure 10.

TABLE A-8  
Level of Contamination on the Surface of a Stainless Steel Table While Exposed in a Laminar Crossflow Room  
(values in viable particles per square foot)

| Sampling Time<br>(in hours) | UPSTREAM OF PERSONNEL                       |                              |                              |  | DOWNSTREAM OF PERSONNEL                     |                              |                              |  | UNOCCUPIED                                  |                              |                              |  |
|-----------------------------|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|--|
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  |
| 0                           | 9   | 32.4                         | 1.09                         |  | 9   | 32.4                         | 1.09                         |  | 0   | 16.6                         | 0                            |  |
| 2                           | 0   | 16.6                         | 0                            |  | 36  | 71                           | 15.5                         |  | 0   | 16.6                         | 0                            |  |
| 4                           | 0   | 15.6                         | 0                            |  | 36  | 71                           | 15.5                         |  | 4.5   | 25                           | 0.114                        |  |
| 6                           | 4.5   | 25                           | 0.11                         |  | 40  | 77                           | 18.5                         |  | 13.5  | 39.4                         | 3                            |  |
| 23                          |   |                              |                              |  |   |                              |                              |  | 18  | 46                           | 4.5                          |  |

Note: Table corresponds to Figure 11.

TABLE A-9  
Microbial Fallout on Blood Agar Plates in a Laminar Downflow Room  
(values in viable particles per square foot)

| Sampling Time<br>(in hours) | UNOCCUPIED                                  |                                   |                                   |   |                                   |                                   | OCCUPIED                                    |                                   |                                   |
|-----------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|---|-----------------------------------|-----------------------------------|
|                             | AIR OFF                                     |                                   |                                   | AIR ON                                      |                                   |                                   | AIR ON                                      |                                   |                                   |
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confi-<br>dence<br>Level | Lower<br>Confi-<br>dence<br>Level |
| 2                           | 204   | 224                               | 185                               | 0   | 2                                 | 0                                 | 0   | 2                                 | 0                                 |
| 4                           | 646   | 665                               | 610                               | 0   | 2                                 | 0                                 | 0   | 2                                 | 0                                 |

Note: Table corresponds to Figure 12.

TABLE A-10

Microbial Fallout on Blood Agar Plates in a Laminar Crossflow Room

(values in viable particles per square foot)

| Sampling Time<br>(in hours) | PERSONNEL UPSTREAM                          |                              |                              |  | PERSONNEL DOWNSTREAM                        |                              |                              |  | UNOCCUPIED                                  |                              |                              |
|-----------------------------|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|--|---|------------------------------|------------------------------|
|                             | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |  | Viable<br>Particles<br>per ft. <sup>2</sup> | Upper<br>Confidence<br>Level | Lower<br>Confidence<br>Level |
| 2                           | 5   | 11.6                         | 1.6                          |  | 62  | 76                           | 38                           |  | 0   | 3.7                          | 0                            |
| 4                           | 3   | 8.77                         | 0.62                         |  | 61  | 76                           | 46                           |  | 6   | 13                           | 2                            |

Note: Table corresponds to Figure 13.

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